33. (new) The method of claim 32 wherein the viral vector is a replication deficient adenoviral vector and the cell is a producer cell capable of complementing the deleted functions of the replication deficient adenoviral vector.

34. (new) The method of claim 33 wherein the replication deficient adenoviral vector lacks a functional E1 region and the producer cell is a 293 cell.

35. (new) The method of claim 32 wherein said *in vitro* practice of the method is in a process to purge tumor cells from a stem cell product by exposing said stem cell product to a calpain inhibitor prior to the administration of a viral vector.

36. (new) The method of claim 35 wherein said viral vector is an adenoviral vector that encodes and expresses the p53 tumor suppressor gene.

STATUS OF THE CLAIMS FOLLOWING AMENDMENT

Subject to the entry of the foregoing Amendment, Claims 5-7 and 21-36 are pending in this application. Claims 1-4, 8-16 and 18-20 were cancelled without prejudice in the Amendment filed July 21, 2000 subject to a Requirement for Restriction. The Office Action indicates that the pending Claims 5-7 are free of the Prior Art (Office Action at page 7). However, Claims 5-7 stand rejected pursuant to 35 USC 112, first paragraph, as not being reasonably supported by the teaching of the specification. Claims 21-36 have not been examined, but depend from pending claims which have been examined. No claim is allowed.

REMARKS

I. Remarks Regarding the Amendments to the Claims and New Claims 21-36:

A. Amendment to Claims 5 and 7:

Regarding the Amendment to Claim 5, this is in response to the Examiner's that the present invention is enabling only for the use of micro-calpain inhibitors. Claim 7 was amended to have proper antecedent basis based on the amendment to claim 5.

B. Cancellation of Claim 17:

Claim 17 was cancelled in response to the Examiner's observation that it was duplicative of the pending claim 7 and would, if claim 7 were found allowable, result in a double-patenting

situation. Applicants believe that the foregoing amendment canceling claim 17 obviates this potential problem. Applicants regret this oversight and thank the Examiner for bringing this to the Applicants' attention.

C. Addition of New Claims 21-36:

The foregoing Amendment by the addition of new claims 21-36 does not introduce new matter to the specification. The subject matter of new claims 21-36 is fully supported by the teaching of the specification. Support for the use of both replication deficient (Claim 21, *et seq.*) and replication competent vectors (Claim 28, *et seq.*) is found at page 13, line 31 through page 14, line 10 of the specification. The potential that the vector may encode a therapeutic transgene (Claim 23) is found at page 23, lines 26-34. That the therapeutic transgene may be a cytoxic or pro-apototic transgene (Claims 23, *et seq.*) is described at page 24 lines 24-30 and page 25, line 34 through page 26 line 2 of the specification, respectively. Regarding claim 25, p21 is specifically provided as an example of a cytostatic gene at page 24, line 26. Regarding claims 27 and 31, p53 and E3 11.6K are specifically provided as examples of pro-apoptotic genes at page 26, lines 1-2. The specification specifically describes the practice of the methods *in vivo* or *in vitro* (see pages 27-29) and the Examiner has conceded the specification is enabling for the practice of the present invention in vitro (Office Action at page 2). Replication deficient adenoviral vectors and the need for complementing cell lines for their propagation (Claim 33) is specifically described at page 13, lines 31-34. The specific example of an E1 deleted replication deficient adenoviral vector and the use of the 293 producer cell line (Claim 34) is described in the paragraph bridging pages 13 and 14 of the specification. The use of calpain inhibitors in conjunction with conventional stem cell purging protocols (Claim 35), particularly involving the use of adenoviral vectors (Claim 36) is described in the specification at page 38, lines 3-34. Consequently, Applicants believe that the subject matter of the new claims introduces no new matter to the specification. The addition of these new claims at this time should not pose any significant additional burden on the Examiner as these new claims are dependent from pending claims 5 and 6 which have been examined and do not enlarge the scope thereof.

Applicants therefore believe that entry of the foregoing Amendment is proper and request entry of the Amendment.

II. Claim Rejections – 35 USC 112:

The Office Action indicates that the pending claims are free of the prior art and that the only remaining issues relate to the scope of the pending claims in light of the teaching of the specification. The Examiner states that the pending claims contemplate the practice of the invention *in vitro* and *in vivo*. The Examiner states that the present claims 5-7 are enabled by the specification only if they are amended to relate to the practice of the invention *in vitro* and if they are restricted to the use micro-calpain inhibitors. The Examiner states that the specification "while enabling for an *in vitro* method of increasing the infectivity of a cell to a viral vector by treatment of the cell with a micro-calpain inhibitor," it does not reasonably provide enablement for the practice of the claimed method *in vivo*. In view of the foregoing amendment to Claim 5 (from which all pending claims depend) wherein the calpain inhibitor is specified as being a micro-calpain inhibitor, the Applicants believe that the rejection of the claims regarding the scope of calpain inhibitors contemplated is obviated.

The remaining issue regarding the allowability of the pending claims relates to whether the claims are enabled for the practice of the present method *in vivo*. The Examiner makes two primary arguments in this regard:

(1) that there is insufficient teaching to enable the skilled artisan to practice the claimed invention based on the lack of predictability in the field of gene therapy, and

(2) there is insufficient scientific data presented to demonstrate that the invention will work *in vivo*.

The Applicants traverse.

A. The Teaching of the Specification Does Provide Sufficient Guidance to the Skilled Artisan to Practice the Method of the Present Invention.

The specification provides specific guidance relating to the use of calpain inhibitors in vivo. See page 36-37. The level of guidance provided by the specification is sufficient for those of skill in the art to which the invention is directed to practice the invention *in vivo*. As indicated in the specification, the treatment of individuals with viral vectors is known to those of skill in the art. Simply labeling the use of recombinant vectors for therapeutic applications as "gene therapy" does not eviscerate the extensive experience in the field with the use of viral vectors. Individuals have been treated with viral vector vaccines for more than 50 years and their routes

of administration, pharmacokinetics and pharmacodynamics are well established. Similarly, viral vectors have been used for many years in the treatment of human beings while more recent studies have been employed with recombinantly modified viral vectors. Reports date from the mid-1950s on the use of viral vectors for the treatment of cervical cancer with many other physician initiated clinical applications of viral vectors reported in the scientific literature. Although the level of success achieved with such treatments is difficult to determine by current standards, what is important is that the skilled artisan was able to sufficiently formulate and administer a viral vector to a human being without untoward side effects more than 40 years ago. Given the recent extensive experience in this field where gene therapy agents are entering Phase III clinical trials (*e.g.* ONYX-015/dl1520 is currently entering Phase III clinical trials in the United States), it would certainly be within the scope of the skilled artisan to use a micro-calpain inhibitor *in vivo* based on the data presented here and that which is available to those of skill in the art.

As indicated, the present invention provides a means for enhancing the transduction efficiency of a viral vector by approximately 100 fold at a concentration of 10 micromolar. It would be readily apparent to those of skill in the art to adjust the dosage of the vector employed based on the concentration of micro-calpain inhibitor used to effect the equivalent therapeutic effect as the full dose in the absence of a micro-calpain inhibitor. A discussion of the formulation of this agent in combination with viral vectors is described at page 32-34 of the specification. The specification makes specific reference to the concentration and dosage regimens employed with gene therapy vectors. Clearly, the amount of calpain inhibitor to be employed will vary depending on the quantity of the virus used, the route of administration, and the pharmacological properties of the virus. However, as is discussed in more detail below, the present invention is not claiming a method of treating a human being with a viral vector but rather enhancing the infectivity of the cells to the viral vector. Whether or not the viral vector employed provides a therapeutic benefit to the individual is collateral to the patentability of the present invention.

B. The Rejection of the Claims is Improper Because the Examiner has Improperly Interpreted the Scope of the Claims.

While the pending claims are potentially encompass both the practice of the present method *in vivo* and *in vitro*, the Examiner has expanded the claim of the present invention to include the requirement not only that the calpain inhibitors enhance the infectivity of a cell to a

viral vector *in vitro* and *in vivo*, but that the specification demonstrate an increase in transduction efficiency resulting in an enhanced therapeutic benefit to a human being from the vector being administered. At page 4 of the Office Action, the Examiner states:

> The claimed method is an adjunct to gene therapy. Gene therapy and adjuncts to gene therapy are not routinely successful. Therefore, the disclosure must enable the full scope of the claimed methods with specific guidance. However, the specification is not enabling for in vivo applications of the claimed method. At the time the application was filed, the art of administering any type of genetic expression vector to an individual **so as to provide a tangible therapeutic benefit was poorly developed and unpredictable.**

This is clearly an improper expansion of the present claims. The pending claims are directed to a method enhancing the infectivity of a cell to a viral vector by the use of a calpain inhibitor. As long as ability of the micro-calpain inhibitors to increase in the infectivity of a cell to a viral vector is supported by the specification, that is the end of the inquiry pursuant to 35 USC 112, first paragraph. Whether or not the subject receives a therapeutic benefit from the viral vector to be administered is irrelevant to the scope of the pending claims. Consequently, the alleged failure of gene therapy vectors to provide a "tangible therapeutic benefit" is simply not relevant to the rejection of the pending claims pursuant to 25 USC 112, first paragraph.

C. The Rejection of the Pending Claims Pursuant to 35 USC 112, First Paragraph is Improper Because the Examiner has Not Addressed the Primary Requirement of 35 USC 112, First Paragraph, but Rather the Utility Requirement of 35 USC 101.

There is a fundamental distinction between a rejection based on a lack of utility pursuant to 35 USC 101 and the failure to comply with the requirements of 35 USC 112, first paragraph. If the Examiner is questioning whether or not the invention works as claimed, the rejection which is proper is based on 35 USC 101. However, a rejection based on 35 USC 112 first paragraph is proper where one of skill in the art would need information in addition to the specification and that available to the skilled artisan to practice the invention. Consequently, the question for resolution is whether or not the specification has provided those of skill in the art with the requisite guidance to practice the invention and not whether the invention works as claimed. An analysis of the Examiner's reasons in support of the rejection of claims of claims 5-7 pursuant to 35 USC 112 first paragraph will demonstrate the impropriety of rejection of the pending claims as what the Examiner is truly questioning is whether or not mechanism of action of the agent *in vivo* is the same as the mechanism of action *in vitro*.

Although the Examiner has conceded that claims directed to the *in vitro* practice of the method of the present invention with a micro-calpain inhibitor are enabled[1], the Examiner maintains that the lack of scientific data fails to demonstrate that the present invention will function similarly *in vivo*. The Examiner alleges that this enablement for the *in vivo* practice of the presently claimed method is not provided by the specification due to the lack of working examples of the invention *in vivo*. Although the Applicants have provided a working example of the enhanced anti-tumor effect *in vivo* in a xenograft immunocompetent murine tumor model wherein the tumor growth was slowed more in the presence of rAd-p53 and CI-1 as compared to each agent alone. The Examiner states that this *in vivo* data is unpersuasive for two reasons: (1) that it is impossible to determine whether the enhanced anti-tumor effect is due to enhanced infectivity or merely the cumulative effect of each agent which possesses anti-tumor efficacy, and (2) that the data is impossible to interpret due to the labeling of the columns ANCB[2] in Figures 12A, B and C without a description of this term in the specification. Since calpain inhibitor 1 can induce apoptosis alone, the Examiner questions whether the enhanced anti-tumor effect described in Figures 12 A, B and C is due to an enhancement of infectivity of the cells to the virus or merely the cumulative pro-apoptotic effects of the rAd-p53 virus and the calpain inhibitor.

Although it might be scientifically interesting to have data demonstrating that more tumor cells were infected by the virus in the presence of CI-1 in this model to demonstrate that the *in vivo* mechanism of action is the same as that observed *in vitro*, three points ***are*** conclusively demonstrated by the data presented from this murine model:

1. the administration of a calpain inhibitor *in vivo* is having an effect – it is reaching the tumor cells and it is having an anti-tumor effect *in vivo* at the dosage ranges provided in the experiment;

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[1] The Office Action states at page 2, "*...the specification, while being enabling for a in vitro method of increasing the infectivity of a cell to a viral vector by treatment of a cell with a micro-calpain inhibitor, ...*"

[2] ANCB describes a replication deficient recombinant adenovirus expressing a non-secreted form of the alkaline phosphatase gene under control of the constituitive CMV promoter. The ANCB vector is the same as the FTCB vector except that the transgene in the FTCB vector is the p53 gene. Applicants regret this oversight that this information was not provided in the specification, but do not believe that it affects the value of the remaining data presented comparing the anti-tumor effects of FTCB and CI-1 alone and in combination and demonstrating an enhanced effect in combination.

2. the recombinant adenoviral vector encoding p53 is reaching the tumor cells and having an anti-tumor effect; and

3. the combination of CI-1 and the rAd-p53 vector produces an enhanced anti-tumor effect.

The Applicants assert that this enhanced anti-tumor effect is due to the increase in infectivity of the tumor cells in the presence of the micro-calpain inhibitor. This is not merely conjecture but is supported by the *in vitro* studies.

Even under the utility examination guidelines the Examiner's position is not tenable. In the absence of some soundly based reason to question whether or not the invention will work as claimed, the Examiner is bound to accept the Applicants assertion of utility. The Examiner has provided no such reason. The Examiner has not provided any reasons or scientific support why the use of micro-calpains *in vivo* would not be useful. The fact that gene therapy viral vectors have had mixed clinical results is irrelevant. This experience does not cast doubt on functionality of the invention as claimed.

Perhaps what is most important about the data is what it does not provide – it does not provide the Examiner with a reasonable basis to question whether or not the invention works as claimed by the Applicants. Had the data failed to show an enhanced anti-tumor effect *in vivo*, there might be grounds to question whether or not the method of the present invention would indeed be functional *in vivo*. However, this was not the case.

Finally, a demonstration of the mechanism of action at work *in vivo* is not required to satisfy 35 USC 101 and is clearly irrelevant to the question of patentability under 35 USC 112, first paragraph. If such data were provided, it would not enhance the ability of the skilled artisan to practice the claimed invention *in vivo* but would rather simply provide an additional demonstration that the invention worked as asserted *in vivo*. The fact that the Examiner would find such data relevant to the issue demonstrates that what is truly at issue in the mind of the Examiner is whether or not the invention works *in vivo*, not whether the specification provides guidance to those of skill in the art to practice the invention *in vivo* as required by 35 USC 112, first paragraph.

Since the Examiner has provided no scientific evidence to the contrary, the Applicants assertion that the invention works as claimed must be accepted. The Applicants believe that

based on the teaching of the specification in conjunction with the information available to those of skill in the art regarding the use of viral vectors *in vivo*, it would not require undue experimentation for the ordinarily skilled artisan to use this agent in conjunction with the administration of a viral vectors to achieve an enhancement in viral infectivity of the target cells *in vivo*. The Examiner's improper characterization of the present invention as "gene therapy" coupled with the fact that some rather outdated review articles cited by the Examiner question the therapeutic benefit of gene therapy viral vectors does not make the present invention "unpredictable" requiring some greater level of disclosure. Given the extensive experience with the administration of viral vectors in vivo and the quantitated increase in infectivity relative to calpain inhibitor concentration provided by the specificaiton, the ordinarily skilled artisan would readily be able to practice the present method in vivo.

The Applicants therefore believe that the rejection of claims 5-7 pursuant to 35 USC 112, first paragraph is in error and respectfully request that it be withdrawn. Since new pending claims 21-36 are dependent from this claims, the Applicants believe that the new claims are similarly without basis of rejection and assert that they are similarly patentable

CONCLUSION

Based on the foregoing reasons of fact and law, the Applicants that the pending claims, as amended are in condition for allowance. Consequently, the Applicants respectfully request that this pending claims in this application be granted favorable consideration and this application passed to issuance without further delay.

If the Examiner believes that an interview would expedite the prosecution of this application, the Applicants' attorney would welcome the opportunity to discuss this application further with the Examiner by telephone to resolve any outstanding issues.

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Respectfully submitted,

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